PCT

WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

BB

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 7 :
A61K 49/00

A2

(11) International Publication Number: WO 00/35490

(43) International Publication Date: 22 June 2000 (22.06.00)

(21) International Application Number: PCT/EP99/09783

(22) International Filing Date: 10 December 1999 (10.12.99)

(30) Priority Data:
9804328–4 15 December 1998 (15.12.98) SE

(71)(72) Applicant and Inventor: BUSCH, Christer [SE/SE]; Nya Valsättravägen 17, S–756 46 Uppsala (SE).

(74) Agents: NILSSON, Brita et al.; AB Stockholms Patentbyrå, Zacco & Bruhn, P.O. Box 23101, S–104 35 Stockholm (SE).

(81) Designated States: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
Without international search report and to be republished upon receipt of that report.

(54) Title: CONTRAST AGENT FOR FACILITATING OCULAR IDENTIFICATION AND INSPECTION OF LYMPH NODES

(57) Abstract

A contrast agent, and a method, for facilitating ocular identification and inspection of lymph nodes are disclosed. The contrast agent comprises a non–carcinogenic non–toxic biodegradable conjugate of a macromolecule and a reactive dye. Preferably the macromolecule is selected from hyaluronan, dextrans, glycogens, denaturated albumin molecules, and so–called sulfur colloids, and the reactive dye is selected from triazine based reactive dyes, such as Cibachron dyes or Procion dyes, and pyrimidinyl based reactive dyes, such as Rectone dyes, and reactive alkene dyes, such as Ramazol dyes, patent violet, trypan blue, and isosulfan blue.

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece			TR	Turkey
BG	Bulgaria	HU	Hungary	ML	Mali	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MN	Mongolia	UA	Ukraine
BR	Brazil	IL	Israel	MR	Mauritania	UG	Uganda
BY	Belarus	IS	Iceland	MW	Malawi	US	United States of America
CA	Canada	IT	Italy	MX	Mexico	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NE	Niger	VN	Viet Nam
CG	Congo	KE	Kenya	NL	Netherlands	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NO	Norway	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	NZ	New Zealand		
CM	Cameroon			PL	Poland		
CN	China	KR	Republic of Korea	PT	Portugal		
CU	Cuba	KZ	Kazakstan	RO	Romania		
CZ	Czech Republic	LC	Saint Lucia	RU	Russian Federation		
DE	Germany	LI	Liechtenstein	SD	Sudan		
DK	Denmark	LK	Sri Lanka	SE	Sweden		
EE	Estonia	LR	Liberia	SG	Singapore		

Contrast agent for facilitating ocular identification and inspection of lymph nodes

The present invention relates to a contrast agent for facilitating ocular identification and inspection of lymph nodes, and to a method of facilitating ocular identification and inspection of lymph nodes.

Background

In cancer surgery, lymphadenectomies are often performed with either or both the aim of eliminating cancer disease which has spread to lymph nodes and staging the disease. The intervention may be made by open surgery or by so-called laparoscopy. The surgeon generally identifies the fat tissue strings which from experience are known to include the lymph nodes and which are located close to the large blood vessels, and above all the veins. The outcome of this type of surgery may vary depending on if the surgeon can identify the single lymph nodes in the fat masses, since these have a color and consistency which are very like those of the fat.

The lymphadectomic preparations are sent to the pathology department, usually for freeze sectioning, i.e. rapid diagnostics, for the determination of if, or not, there is cancer in the lymph nodes by a combination of examination by visual macroscopy and palpation. This is then followed by a so-called cryostatic sectioning, staining and microscopic inspection for the purpose of identifying cancer metastases in the lymph nodes.

Based on the result of this procedure, the subsequent treatment of the patient is decided.

The pathological inspection for localization of the lymphnodes in the fat tissue preparations obtained by resection is often very difficult due to the slight difference in the consistency and color.

There are some techniques disclosed in the prior art which aid the identification of lymph nodes and the staging of cancer. Many of these are based on radiation responsive instruments and radiolabeled locators. (See e.g. Offodile R., et al. Minimally Invasive Breast Carcinoma Staging using Lymphatic Mapping with radiolabeled dextran. Cancer, 1998 May, 82:9, 1704-8; Albertini J.J., et al. Intraoperative Radiolymphoscintigraphy Improves Sentinel Lymph Node Identification for Patients with Melanoma. Annals of Surgery, 1996, Vol. 223, No. 2, 217-225.) Another approach to visualize lymph nodes was injection of a vital dye at the primary breast cancer site, and the axillary incision was standardized to approximately 5 minutes after the injection. (Guiliano A. E., et al. Lymphatic Mapping and Sentinel Lymphadenectomy for Breast Cancer. Annals of Surgery, 1994, Vol. 220, No. 3, 391-401.)

Description of the invention

The present invention provides a non-radioactive contrast agent, and a method, for facilitating ocular identification and inspection of lymph nodes. The contrast agent comprises a conjugate of a macromolecule and a reactive dye. The conjugate is arrested by phagocytic cells in the lymph nodes and is kept there by virtue of the macromolecule while the dye provides the coloring. The macromolecule prolongs the duration of the conjugate in the lymph nodes in relation to unconjugated dye molecules, which pass pretty quickly through the lymph nodes after a short accumulation therein.

The conjugate of the invention may be injected interstitially to an area close to lymph nodes of interest in an individual, i.e. primarily to an area of a primary cancer. The conjugate is then transported by the lymphatic tract to lymph nodes and is arrested and accumulated therein. The time interval between the injection and the incision may vary depending on the type and size of the conjugate, but will preferably be in the range of 30 min to 2 days.
The conjugate of the invention is preferably water-soluble or colloidal, and it is injected in the form of a solution or suspension in a physiologically acceptable vehicle, such as saline.

Thus, one aspect of the invention is directed to a contrast agent for facilitating ocular identification and inspection of lymph nodes, which comprises a non-carcinogenic non-toxic biodegradable conjugate of a macromolecule and a reactive dye.

In an embodiment of this aspect of the invention the macromolecule is selected from macromolecules known to undergo receptor mediated phagocytos.

In another embodiment the conjugate has a large enough size to be arrested by the sinusoidal histiocytes of lymph nodes.

In a preferred embodiment the macromolecule is selected from the group consisting of hyaluronan, dextrans, glycogens, denaturated albumin molecules, and so-called sulfur colloids, and the reactive dye is selected from the group consisting of triazine based reactive dyes, such as Cibachron dyes or Procion dyes, and pyrimidinyl based reactive dyes, such as Rectone dyes, and reactive alkene dyes, such as Ramazol dyes, patent violet, trypan blue, and isosulfan blue.

In a most preferred embodiment the macromolecule is a dextran having a molecular weight (weight average molecular weight) of from 1 000 to 500 000, preferably from 10 000 to100 000, and the reactive dye is Cibachron Blue F3G-A, Ciba Brilliant R 2Gp or Reactonmarin blau GRL.

In a further preferred embodiment the degree of substitution expressed as mmol of dye per gram of conjugate is between 0.01 - 1.0, preferably between 0.05 and 0.5.

Another aspect of the invention is directed to a method of facilitating ocular identification and inspection of lymph nodes comprising interstitial injection, to an area close to lymph nodes of interest in an individual, of a solution or suspension of a contrast agent, which contrast agent comprises a non-carcinogenic non-toxic biodegradable conjugate of a macromolecule and a reactive dye, in an amount providing a color to the lymph nodes which are visible to the naked eye after permitting the conjugate to localize in the lymph nodes and after incision.

The individual referred to may be an animal or human patient.

Also in this aspect of the invention, preferably the macromolecule is selected from macromolecules known to undergo receptor mediated phagocytos or the conjugate has a large enough size to be arrested by the sinusoidal histiocytes of lymph nodes.

Further, preferably the macromolecule is selected from the group consisting of hyaluronan, dextrans, glycogens, denaturated albumin molecules, and so-called sulfur colloids, and the reactive dye is selected from the group consisting of triazine based reactive dyes, such as Cibachron dyes or Procion dyes, and pyrimidinyl based reactive dyes, such as Rectone dyes, and reactive alkene dyes, such as Ramazol dyes, patent violet, trypan blue, and isosulfan blue. Preferably the macromolecule is a dextran having a molecular weight of from 1 000 to 500 000, and the reactive dye is Cibachron Blue F3G-A, Ciba Brilliant R 2Gp or Reactonmarin blau GRL, and preferably the degree of substitution expressed as mmol of dye per gram of conjugate is between 0.01 - 1.0.

The invention will now be illustrated by the following examples and experiments, but these should not be considered limiting to the scope of the invention defined in the claims.

Preparation of conjugates

Derivatives of dextran with dyes

J.F.Kennedy (Advan.Carbohydr.Chem.) teaches that commercial triazinebased reactive dyes react with polysaccharides to form stable coloured derivatives. Blue dextran 2000 (Amersham Pharmacia Biotech, Uppsala, Sweden) is a water-soluble derivative of dextran prepared from Cibachron Blue.

Example 1. Blue dextran 70

Dextran T70 (6 g; Amersham Pharmacia Biotech, Uppsala, Sweden) is dissolved in water (25 ml). Cibachron blue F3 G-A, (0.6 g; Ciba-Geigy, V. Frölunda, Sweden) is added with stirring followed by 0. 15 ml 50%(w/v) sodium hydroxide. The mixture is stirred at 40°C overnight and precipitated by adding 99% ethanol (75 ml) . The supernatant is decanted and the residue is redissolved in water (50 ml), neutralised with dilute hydrochloric acid and

precipitated by adding ethanol (150 ml). The reprecipitation is repeated until a clear supernatant is obtained and the product is free from non-bound dye. The product is obtained as a powder by redissolving the residue in water (50 ml) and precipitating in ethanol (500 ml). The blue precipitate is filtered and dried in vacuo at 50°C.

Yield, 5.1 g

Substitution; 0.08 mmol Cibablue/g blue dextran

Example 2. Blue dextran 70 (high substitution)

Dextran T70 (10 g; Amersham Pharmacia Biotech) is dissolved in water (150 ml). Cibachron blue F3 G-A(2.6 g) is added with stirring followed by 4 ml 6N sodium hydroxide. The mixture is stirred at 40°C overnight and precipitated by adding 99% ethanol (250 ml) . The supernatant is decanted and the residue is redissolved in water (150 ml), neutralized with dilute hydrochloric acid and precipitated by adding ethanol (250 ml). The reprecipitation is repeated until a clear supernatant is obtained and the product is free from non-bound dye. The product is obtained as a powder by redissolving the residue in water (100 ml) and precipitating in ethanol (1.5 L). The blue precipitate is filtered and dried in vacuo at 50°C.

Yield, 9.6 g

Substitution; 0.19 mmol Cibablue/g blue dextran

Example 3. Blue dextran 10

Dextran T10 (10 g; Amersham Pharmacia Biotech) is dissolved in water (150 ml). Cibachron blue F3 G-A(1.0 g) is added with stirring followed by 4 ml 6N sodium hydroxide. The mixture is stirred at 40°C overnight and precipitated by adding 99% ethanol (500 ml) . The supernatant is decanted and the residue is redissolved in water (100 ml), neutralised with dilute hydrochloric acid and precipitated by adding ethanol (300 ml). The reprecipitation is repeated until a clear supernatant is obtained and the product is free from non-bound dye. The product is obtained as a powder by redissolving the residue in water (100 ml) and precipitating in ethanol (1 L). The blue precipitate is filtered and dried in vacuo at 50°C.

Yield, 9.2 g

Substitution; 0.1 mmol Cibablue/g blue dextran

Example 4. Blue dextran 40

Dextran T40 (10 g; Amersham Pharmacia Biotech) is dissolved in water (150 ml). Cibachron blue F3 G-A (1.0 g) is added with stirring followed by 4 ml 6N sodium hydroxide. The mixture is stirred at 40°C overnight and precipitated by adding 99% ethanol (500 ml) . The supernatant is decanted and the residue is redissolved in water (100 ml), neutralised with dilute hydrochloric acid and precipitated by adding ethanol (300 ml). The reprecipitation is

repeated until a clear supernatant is obtained and the product is free from non-bound dye. The product is obtained as a powder by redissolving the residue in water (100 ml) and precipitating in ethanol (1 L). The blue precipitate is filtered and dried in vacuo at 50°C.

Yield, 9.1 g

Substitution; 0.11 mmol Cibablue/g blue dextran

Example 5. Blue dextran 70

Dextran T70 (6 g; Amersham Pharmacia Biotech) is dissolved in water (30 ml). Reactonmarin blau GRL (0.6 g; Ciba-Geigy, V.Frölunda, Sweden) is added with stirring followed by 0.15 ml 50%(w/v) sodium hydroxide. The mixture is stirred at 40°C overnight and precipitated by adding 99% ethanol (75 ml) . The supernatant is decanted and the residue is redissolved in water (50 ml), neutralised with dilute hydrochloric acid and precipitated by adding ethanol (150 ml). The reprecipitation is repeated until a clear supernatant is obtained and the product is free from non-bound dye. The product is obtained as a powder by redissolving the residue in water (50 ml) and precipitating in ethanol (500 ml). The blue precipitate is filtered and dried in vacuo at 50°C.

Yield, 3.9 g

Substitution; 0.06 mmol Reactonmarin blau/g blue dextran

Example 6. Red dextran 70

Dextran T70 (10 g; Amersham Pharmacia Biotech) is dissolved in water (150 ml). Ciba Brilliant R 2GP (1.0 g; Ciba-Geigy, V.Frölunda, Sweden) is added with stirring followed by 4 ml 6N sodium hydroxide. The mixture is stirred at 40°C overnight and precipitated by adding 99% ethanol (500 ml) . The supernatant is decanted and the residue is redissolved in water (100 ml), neutralised with dilute hydrochloric acid and precipitated by adding ethanol (300 ml). The reprecipitation is repeated until a clear supernatant is obtained and the product is free from non-bound dye. The product is obtained as a powder by redissolving the residue in water (100 ml) and precipitating in ethanol (1 L). The red precipitate is filtered and dried in vacuo at 50°C. Yield, 9.9 g

Experiments

The following experiments illustrates the usefulness of the invention.

Experiment 1

Animal used in the experiment: Pig, 20 kg

A nearly saturated aqueous solution of Blue dextran 70 (Example 2), 0.25 ml per kg body weight was injected into the groin. Three hours later, the animal was sacrificed, and regional lymph nodes in the lower part of pelvis were identified and found to be blue.

Experiment 2

Animal used in the experiment: Mouse 50 g

A nearly saturated aqueous solution of Blue dextran 70 (Example 2), 0.25 ml per kg body weight was injected into the root of the tail. The next day, the mouse was sacrificed, and regional lymph nodes were identified and found to be blue.

Claims

1. Contrast agent for facilitating ocular identification and inspection of lymph nodes, which comprises a non-carcinogenic non-toxic biodegradable conjugate of a macromolecule and a reactive dye.

2. Contrast agent according to claim 1, wherein the macromolecule is selected from macromolecules known to undergo receptor mediated phagocytos.

3. Contrast agent according to claim1, wherein the conjugate has a large enough size to be arrested by the sinusoidal histiocytes of lymph nodes.

4. Contrast agent according to claim1, wherein the macromolecule is selected from the group consisting of hyaluronan, dextrans, glycogens, denaturated albumin molecules, and so-called sulfur colloids, and the reactive dye is selected from the group consisting of triazine based reactive dyes, such as Cibachron dyes or Procion dyes, and pyrimidinyl based reactive dyes, such as Rectone dyes, and reactive alkene dyes, such as Ramazol dyes, patent violet, trypan blue, and isosulfan blue.

5. Contrast agent according to claim 4, wherein the macromolecule is a dextran having a molecular weight of from 1 000 to 500 000, and the reactive dye is Cibachron Blue F3G-A, Ciba Brilliant R 2Gp or Reactonmarin blau GRL.

6. Contrast agent according to claim 4 or 5, wherein the degree of substitution expressed as mmol of dye per gram of conjugate is between 0.01 - 1.0.

7. Method of facilitating ocular identification and inspection of lymph nodes comprising interstitial injection, to an area close to lymph nodes of interest in an individual, of a solution or suspension of a contrast agent, which contrast agent comprises a non-carcinogenic non-toxic biodegradable conjugate of a macromolecule and a reactive dye, in an amount providing a color to the lymph nodes which are visible to the naked eye after permitting the conjugate to localize in the lymph nodes and after incision.

8. Method according to claim 7, wherein the macromolecule is selected from macromolecules known to undergo receptor mediated phagocytos.

9. Method according to claim 7, wherein the conjugate has a large enough size to be arrested by the sinusoidal histiocytes of lymph nodes.

10. Method according to claim 7, wherein the macromolecule is selected from the group consisting of hyaluronan, dextrans, glycogens, denaturated albumin molecules, and

so-called sulfur colloids, and the reactive dye is selected from the group consisting of triazine based reactive dyes, such as Cibachron dyes or Procion dyes, and pyrimidinyl based reactive dyes, such as Rectone dyes, and reactive alkene dyes, such as Ramazol dyes, patent violet, trypan blue, and isosulfan blue.

11. Method according to claim10, wherein the macromolecule is a dextran having a molecular weight of from 1 000 to 500 000, and the reactive dye is Cibachron Blue F3G-A, Ciba Brilliant R 2Gp or Reactonmarin blau GRL.

12. Method according to claim 10 or 11, wherein the degree of substitution expressed as mmol of dye per gram of conjugate is between 0.01 - 1.0.

PCT WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 7 : A61K 49/00 | A3

(11) International Publication Number: **WO 00/35490**

(43) International Publication Date: 22 June 2000 (22.06.00)

(21) International Application Number: PCT/EP99/09783

(22) International Filing Date: 10 December 1999 (10.12.99)

(30) Priority Data:
9804328-4 15 December 1998 (15.12.98) SE

(71)(72) Applicant and Inventor: BUSCH, Christer [SE/SE]; Nya Valsättravägen 17, S-756 46 Uppsala (SE).

(74) Agents: NILSSON, Brita et al.; AB Stockholms Patentbyrå, Zacco & Bruhn, P.O. Box 23101, S-104 35 Stockholm (SE).

(81) Designated States: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
With international search report.

(88) Date of publication of the international search report:
21 September 2000 (21.09.00)

(54) Title: CONTRAST AGENT FOR FACILITATING OCULAR IDENTIFICATION AND INSPECTION OF LYMPH NODES

(57) Abstract

A contrast agent, and a method, for facilitating ocular identification and inspection of lymph nodes are disclosed. The contrast agent comprises a non-carcinogenic non-toxic biodegradable conjugate of a macromolecule and a reactive dye. Preferably the macromolecule is selected from hyaluronan, dextrans, glycogens, denaturated albumin molecules, and so-called sulfur colloids, and the reactive dye is selected from triazine based reactive dyes, such as Cibachron dyes or Procion dyes, and pyrimidinyl based reactive dyes, such as Rectone dyes, and reactive alkene dyes, such as Ramazol dyes, patent violet, trypan blue, and isosulfan blue.

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece			TR	Turkey
BG	Bulgaria	HU	Hungary	ML	Mali	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MN	Mongolia	UA	Ukraine
BR	Brazil	IL	Israel	MR	Mauritania	UG	Uganda
BY	Belarus	IS	Iceland	MW	Malawi	US	United States of America
CA	Canada	IT	Italy	MX	Mexico	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NE	Niger	VN	Viet Nam
CG	Congo	KE	Kenya	NL	Netherlands	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NO	Norway	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	NZ	New Zealand		
CM	Cameroon			PL	Poland		
CN	China	KR	Republic of Korea	PT	Portugal		
CU	Cuba	KZ	Kazakstan	RO	Romania		
CZ	Czech Republic	LC	Saint Lucia	RU	Russian Federation		
DE	Germany	LI	Liechtenstein	SD	Sudan		
DK	Denmark	LK	Sri Lanka	SE	Sweden		
EE	Estonia	LR	Liberia	SG	Singapore		

INTERNATIONAL SEARCH REPORT

Interna al Application No
PCT/EP 99/09783

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 A61K49/00

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
EPO-Internal WPI

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 98 48845 A (COCKBAIN JULIAN R M ;NYCOMED IMAGING AS (NO); DELECKI DANIEL JOSEP) 5 November 1998 (1998-11-05) the whole document ---	1-12
X	WO 96 17628 A (DIAGNOSTIKFORSCHUNG INST ;LICHA KAI (DE); RIEFKE BJOERN (DE); SEMM) 13 June 1996 (1996-06-13) the claims, page 38, line 6 ---	1-12
X	WO 96 04922 A (DAVIS JOANNE T ;COWARD RODERICK T (CA)) 22 February 1996 (1996-02-22) claims --- -/--	1-3,7-9

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
3 May 2000	15. 06. 2000

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	S. Gustavsson/Eö

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

Interna al Application No
PCT/EP 99/09783

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO 94 21303 A (ALLIANCE PHARMA) 29 September 1994 (1994-09-29) page 2, line 34 - line 35 page 6, line 25 - line 33 ---	1-12
A	DATABASE WPI Section Ch, Week 199524 Derwent Publications Ltd., London, GB; Class B04, AN 1995-183770 XP002900981 & RU 2 020 963 C (MEDICINAL MATERIALS CHEM CENTRE), 15 October 1994 (1994-10-15) abstract ---	1-12
A	DATABASE WPI Section Ch, Week 198009 Derwent Publications Ltd., London, GB; Class B04, AN 1980-15380C XP002900982 & JP 55 007245 A (DAIICHI SEIYAKU CO), 19 January 1980 (1980-01-19) abstract -----	1-12

1

INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP 99/09783

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.: 7-12
because they relate to subject matter not required to be searched by this Authority, namely:

see next sheet

2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).:

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest.

☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP 99/09783

Claims 7-12 relate to methods of treatment of the human or animal body by surgery or by therapy/ diagnostic methods practised on the human or animal body/Rule 39.1.(iv). Nevertheless, a search has been executed for these claims. The search has been based on the alleged effects of the compounds/compositions.

INTERNATIONAL SEARCH REPORT

...rmation on patent family members

Inter nal Application No
PCT/EP 99/09783

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9848845 A	05-11-1998	AU 7221298 A	24-11-1998
		AU 7221398 A	24-11-1998
		AU 7221698 A	24-11-1998
		EP 0979103 A	16-02-2000
		EP 0979107 A	16-02-2000
		WO 9848838 A	05-11-1998
		WO 9848846 A	05-11-1998
WO 9617628 A	13-06-1996	DE 4445065 A	13-06-1996
		AU 709152 B	19-08-1999
		AU 3740995 A	26-06-1996
		CA 2205906 A	13-06-1996
		CN 1174511 A	25-02-1998
		EP 0796111 A	24-09-1997
		HU 77378 A	28-04-1998
		JP 10510250 T	06-10-1998
		NO 972509 A	02-06-1997
		NZ 294568 A	30-08-1999
		ZA 9509707 A	29-05-1996
WO 9604922 A	22-02-1996	AU 3239895 A	07-03-1996
		CA 2196408 A	22-02-1996
		EP 0774970 A	28-05-1997
		US 5772982 A	30-06-1998
WO 9421303 A	29-09-1994	AU 6365894 A	11-10-1994
		US 5460800 A	24-10-1995
RU 2020963 C	15-10-1994	NONE	
JP 55007245 A	19-01-1980	JP 1434195 C	07-04-1988
		JP 62042892 B	10-09-1987